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REVIEW OF THE CONTROL OF JAPANESE BEETLES IN ONTARIO

Ontario Pesticides
Advisory Committee

February 15, 1974

SB
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057
1974

Ontario
Ministry
of the
Environment

The Honourable
William G. Newman,
Minister

Everett Biggs,
Deputy Minister

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JAPANESE BEETLES

Adults of the Japanese beetle usually emerge in Ontario during the first two weeks of July. As adults, they live for 30 to 45 days and feed on a wide variety of plants. After mating, the female will crawl into the soil 1 - 4" deep, and lay her eggs usually one to four in a given location. Each female will lay 40 to 60 eggs. Larvae hatch in two to three weeks and the grubs go through three instars before winter. During these three larval instars, the grubs feed on the roots of grasses in the top three inches of soil. As the soil temperature drops below 60° F, the larvae will migrate downward and pass the winter below the frost line. Thus the population is largely first instar larvae in late August and early September; second instar larvae late September, early October; third instar larvae in late October, early November. As the soil temperature warms in the spring, the third instar larvae approach the soil surface again and do a significant amount of feeding before transferring to the perpupal stage, then to pupae, and later emerging as adults.

The Japanese beetle was first recorded in the United States in 1916. It soon spread rapidly throughout the eastern and north central part of the United States and has been present in Canada since the early 1930's. The Division of Plant Protection has conducted trapping for the Japanese beetle since 1935 and soil treatments for control (eradication) of larvae have been carried out since 1941. In the early days, the treatment used was lead arsenate at the rate of 500 lbs per acre. From 1949 to 1955, 50% wettable DDT was used at the rate of 50 lbs per acre. From 1956 to 1965, dieldrin was used at the rate of 30 lbs per acre of 10% granular material. Since that time, chlordane has been used at the rate of 8 lbs active ingredient per acre. The insect has been present in many areas in Ontario and in addition, has been reported from Quebec and Halifax, Nova Scotia. Treatments in Ontario have been conducted jointly by the Division of Plant Protection, Ottawa, and the Farm Products Inspection Branch, Ontario Ministry of Agriculture and Food. Acreages treated each year have been relatively small as indicated in Table 1 for the period 1966 through 1972.

Table 1. Treatment for eradication of the Japanese beetle, 1966-72

Location	Years	Acres
Rockport, Ivylea	1968; 70; 72	42
Belleville	1970	100
St. Catherines	1966; 67; 68	125
Niagara Falls	1967; 68	1000
Fort Erie	1966; 67	115
Dunnville	1971; 72	65
Hamilton	1966	200
Windsor	1966	10

Treatment - chlordane, 8 lbs active ingredient per acre.

Treatments for eradication of the Japanese beetle have normally been made in October.

Research work on control of the Japanese beetle has been carried out largely in New York State and in Ohio. In these areas, it was learned that the cyclodiene insecticides: chlordane, aldrin, dieldrin, and heptachlor, provided effective control for Japanese beetle larvae. Tests in New York indicated that an application of chlordane at 10 lbs. active ingredient per acre or dieldrin at 3 lbs active ingredient per acre would provide control for at least six years. More recently, Tashiro and Fiori (1969), conducted tests which convinced them that the rate of application for either of these compounds could be reduced by a factor of three and satisfactory control still be obtained. In 1972 Tashiro and Neuhauser discovered a population of Japanese beetles in New York State in Onondaga County that were resistant to chlordane (Tashiro and Neuhauser), 1973). In the same year, Japanese beetles resistant to the cyclodiene insecticides were also found in Ohio (Niemczyk and Lawrence, 1973). In both the New York and Ohio studies, comparisons were made between the resistant Japanese beetles and susceptible populations. In each case it was demonstrated that the level of resistance was very high and that control with chlordane was no longer feasible. The Ohio study also reported on the toxicity of related cyclodiene insecticides: aldrin, heptachlor and dieldrin, to adults of the resistant and susceptible strains of the Japanese beetle. Their data showed that those insects resistant to chlordane were also resistant to the other cyclodiene insecticides. This work was extended in 1973 and tests were made on the larval stages with several cyclodiene insecticides. These tests confirmed that insects resistant to chlordane could not be controlled with dieldrin.

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Chlordane-Resistant Japanese Beetle in New York

H. Tashiro[1] and W. Neuhauser[2]

ABSTRACT

The presence of a chlordane-resistant Japanese beetle strain in Liverpool, Onondaga County, New York was established during the 1972 season.

In laboratory tests at 25 C, holding third instar larvae in insecticide-treated soil indicated the presence of resistance also to dieldrin but not to lindane. Holding larvae in soil containing 64 ppm chlordane (approximately equivalent to 64 pounds AI/3-in acre) did not cause 100 per cent mortality in 4 weeks, the action expected with 4 ppm in susceptible larvae during this interval.

Comparison of a susceptible to the resistant first instar larvae in their response to chlordane-treated soil revealed a sixtyfold resistance factor.

Beetles were collected from various localities to obtain eggs for determining insecticidal action on their first instar larvae. The resistant strain was prevalent within 0-2 miles of the original site where resistant larvae were first found. Occurrence of the resistant strain beyond this radius should be suspected.

Insecticidal applications to turf at the original site during late April 1972 followed by larval surveys at 4 and 6.5 weeks post-treatment intervals also revealed that chlordane was not killing the larvae.

INTRODUCTION

Chlordane was first used for control of larvae of the Japanese beetle *Popilla japonica* Newman in turf in 1947 (2). In subsequent years, other chlorinated hydrocarbon insecticides; namely, aldrin, dieldrin, and heptachlor, were recommended for control of this and other scarabaeid larvae in turf. A single application of any of these materials has provided excellent control for several years. As examples, the European chafer *Amphimallon majalis* Razoumowsky was still being controlled after 7 years when the study was terminated (4). Chlordane and heptachlor have controlled *Phyllophaga* spp. and the northern masked chafer *Cyclocephala borealis* Arrow for 12 years in Ohio (5). The first evidence of a species of Scarabaeidae being resistant to chlorinated hydrocarbon insecticides was discovered in larvae of the European chafer during May 1969 (6).

A complaint was received during early April 1972 from the Liverpool Golf and Country Club (suburb of Syracuse, New York) that repeated applications of chlordane during the past 3 years were not controlling Japanese beetle larvae. We immediately suspected species misidentification with presence of a chlorinated hydrocarbon-resistant European chafer population similar to that occurring in the Rochester area (6). However, upon visiting the golf course and examining the larvae, the presence of a Japanese beetle infestation was verified. The occurrence of a resistant strain of this species was immediately suspected. This would not be surprising in view of the occurrence of the resistant strain of the European chafer (6). Dieldrin and chlordane have been used since early 1960 in Syracuse and much of Onondaga County, New York for European chafer control on lawns.

Field and laboratory studies were conducted during the spring and summer of 1972 which verified the existence of a highly resistant strain. The results of these studies follow.

GENERAL PROCEDURES

Laboratory Tests—During April 1972, third instar larvae of the Japanese beetle were collected from the damaged turf of the golf course and were held in soil

at 2-5 C until needed for laboratory studies. Low concentrations (1-2%) granular or dust formulations of insecticides (except carbaryl 10G) were thoroughly incorporated along with a seed mixture (domestic rye and sweet clover) into moist friable sandy loam passed through ½ inch mesh. One-half cubic foot lots of treated soils were placed in five wooden trays (10.5 x 10.5 x 4 inches ID) and infested with larvae by placing them on the soil surface and permitting them to burrow ir Each tray received 25 or 30 individuals. Examinations for survival were made by sifting the soil at weekly intervals from each tray to count the live larvae and returning them to the same soil for further incubation.

For first instar tests, adults were collected during July and placed over finely screened moist soil in which to deposit eggs. Wild grape leaves were placed on the soil surface of oviposition cages as food for the adults. When mature eggs were available, they were transferred to insecticide-treated soils for hatching and insecticidal action on first instar larvae. Preparation of soils was identical to that used for third instar tests except it was screened through ¼ inch mesh. Most treatments for first instar tests consisted of 10, 1-quart clear plastic containers of soil. An egg was placed in each of 30 depressions of 3 mm diameter on lightly compacted soil and covered with 2-3 cm of the same soil preparation.

All laboratory tests were conducted, including holding beetles for egg production, at a constant temperature of 25 C and day-night cycles of 16-8 hours. Water was added to the soil surface when needed to maintain moist soil.

Field Tests—Plots 20 x 20 feet were established on permanent turf with each treatment replicated five times in randomized complete block design. Preweighed amounts of insecticide were applied to each plot in both directions to insure complete and uniform coverage. Granular formulations were applied with Scott's 2-foot spreaders, and aqueous suspensions were applied with sprinkling cans. Spring applications made on April 28, 1972 were not watered in because of frequent rains. Soil surveys of five separate square foot diggings to a depth of 4-6 inches were made in each plot at two different periods to determine populations of grubs following spring applications.

Summer treatments directed against the new brood of young larvae were superimposed over half of each spring plot when the new brood was 17 per cent eggs, 70 per cent first instar, and 13 per cent second instar larvae. Due to inadequate populations in the untreated plots during October 1972 surveys, no results were obtained from these summer treatments.

RESULTS AND DISCUSSION

Laboratory Tests on Third Instar Larvae—A preliminary test indicated clear evidence of resistance to chlordane. Therefore, larvae were exposed to three chlorinated hydrocarbon insecticides: chlordane, dieldrin, and lindane; three organophosphates: diazinon, chlorpyrifos, and pirimiphos-ethyl; and to the carbamate, carbaryl. There were five trays of 25 larvae per treatment. Japanese beetle larvae of a susceptible strain were not available, and direct comparison between the resistant and a susceptible strain could not be made. However, all previous studies (unpublished data) have indicated that larvae held in chlordane and dieldrin-treated soil at 4 ppm were equally affected as when held in diazinon at the same level. Results shown in Table 1 indicate that all three phosphate insecticides at 2 ppm were much more toxic than chlordane or dieldrin at 4 ppm. Chlordane was the less effective of the two. Evidence of resistance to lindane is questionable, but there appears to be some trend in this direction.

A dose-mortality study was made by introducing larvae from the Liverpool Golf and Country Club into soils containing chlordane at levels of 0, 2, 4, 8, 16, 32, and 64 ppm. Each treatment consisted of five trays of 30 larvae each. The results are shown in Table 2. None of the treatments completely eliminated the larvae. Under the conditions of this test, 100 per cent mortality would have been expected in 4 weeks at both the 2 and 4 ppm rates had the larvae been of a susceptible strain. Instead, only 16 per cent mortality was obtained with 4 ppm chlordane. At 64 ppm, only 81 per cent mortality was obtained in 4 weeks. In contrast, 84 per cent mortality was obtained with 2.09 ppm for the same period in a test conducted by Tashiro and Fiori (8), Table 2. Had the 1969 test been held at 25 C instead of at 21.5 C, the differences between these two tests would, no doubt, be even greater since the rate of insecticidal action is positively correlated with temperature. The results obtained in this dose-mortality test indicate that a substantial degree of resistance has developed in this Liverpool population.

Laboratory Studies of First Instar Larvae—A comparison was made of the toxicity of chlordane to first instar larvae from beetles collected at two different locations: from Waterloo, New York where the population was considered to be susceptible to chlordane, and from the Liverpool Golf and Country Club where resistant third instar larvae were present. After mature eggs of each strain were available, they were placed in moist friable soil containing chlordane at the doses shown in Table 3. After 3 weeks at 25 C, all the eggs had hatched, and the oldest individuals in the untreated soil were second instar. The soil from all the containers was then screened at this time to recover the live larvae. All live individuals were considered normal, affected larvae having been killed

Table 1.—Toxicity of soil incorporated insecticides to third instar Japanese beetle larvae of a suspected chlorinated hydrocarbon resistant strain. 1972.

Insecticides and formulation	PPM AI in soil	% mortality, weeks exposure[a,b]			
		1	2	3	4
Diazinon 2G	2	47 a	97 a	100 a	100 a
Chlorpyrifos 1G	2	64 a	93 a	96 a	98 a
Pirimiphos-ethyl 2.1G	2	16 b	90 a	99 a	100 a
Carbaryl 10G	12	71 a	91 a	97 a	100 a
Lindane 1% dust	2	59 a	71 b	85 b	90 a
Dieldrin 1% dust	4	15 b	55 c	63 b	81 b
Chlordane 1% dust	4	1 bc	37 d	51 c	49 b
Untreated—no. alive from original 125 larvae		106 c	93 e	79 d	59 c

a/ Five trays of 25 larvae/treatment, held at 25 C ambient. Mortalities based on Abbott's formula.

b/ Based on mean recoveries each week, figures for each week followed by the same letter are not significantly different at the 5 per cent level according to Duncan's multiple range test.

Table 2.—Degree of resistance of chlorinated hydrocarbon resistant third instar Japanese beetle larvae to chlordane. 1972.

PPM chlordane in soil	% mortality, weeks exposure[a,b]			
	1	2	3	4
2	0 a	0 a	0 a	6 a
4	0 a	0 a	4 a	16 a
8	4 a	18 b	25 b	33 b
16	12 b	25 b	31 b	32 b
32	17 b	38 b	64 c	75 c
64	34 c	58 c	67 c	81 c
Untreated -no. alive from original 150 larvae	121 a	100 a	81 a	63 a

a/ Five trays of 30 larvae/treatment, held at 25 C ambient. Mortalities based on Abbott's formula.

b/ Based on mean recoveries each week, figures for each week followed by the same letter are not significantly different at the 5 per cent level according to Duncan's multiple range test.

earlier during the young first instar period. Complete kill of the susceptible strain was obtained in soil containing 0.25 ppm chlordane, while it required soils containing 32 ppm chlordane to kill all of the resistant larvae. The 0.25 ppm chlordane required to produce 100 per cent mortality of the susceptible strain compares favorably with results reported by Fleming et al. (3) which indicate that 0.4 pound chlordane per 3-inch acre (approximately equivalent to .4 ppm) was required for 100 per cent mortality. By plotting the information shown in Table 3 on log probit scales to determine LD_{50} values, it was estimated that there was a sixtyfold increase in resistance in the first instar larvae of the Liverpool strain (Fig. 1).

Geographic Distribution of Resistant Strains—Beetles were collected from host plants from 22 different areas of Onondaga County fanning out from the Liverpool Golf Course. Three collections outside of Onondaga County included one from Waterloo in Seneca County, and two from Monroe County. One Monroe County site has a mixed population of Japanese beetle and the chlorinated hydrocarbon-resistant European chafer strain. The other site at Webster Beach Park, adjacent to the resistant area, has a mixed population of the two species, neither

Table 3.--Toxicity of chlordane incorporated soil to chlordane-susceptible and resistant first instar Japanese beetle larvae.

PPM chlordane in soil	Avg no. larvae surviving/container[a]	% mortality[b]
	Susceptible Strain[c]	
0	16.3 b	–
.031	18.3 ab	0
.063	5.7 e	65.0
.125	.3 f	98.2
.25	0 f	100
.5	0	100
1.0	0	100
2.0	0	100
4.0	0	100
	Resistant Strain[d]	
0	22.1 a	–
1	20.6 a	6.8
2	14.9 bc	32.6
4	12.4 cd	43.9
8	9.2 de	58.4
16	1.6 f	92.8
32	0 f	100
64	0	100
128	0	100
256	0	100

a/ Ten containers of 30 eggs/treatment held at 25 C ambient, larval counts made when second instar larvae first appeared in untreated soil. Means followed by the same letter are not significantly different at the 5 per cent level according to Duncan's multiple range test.

b/ Based on Abbott's formula.

c/ Beetles for obtaining eggs collected in Waterloo, Seneca County, New York.

d/ Beetles collected Liverpool Golf Course, Liverpool, Onondaga County, New York.

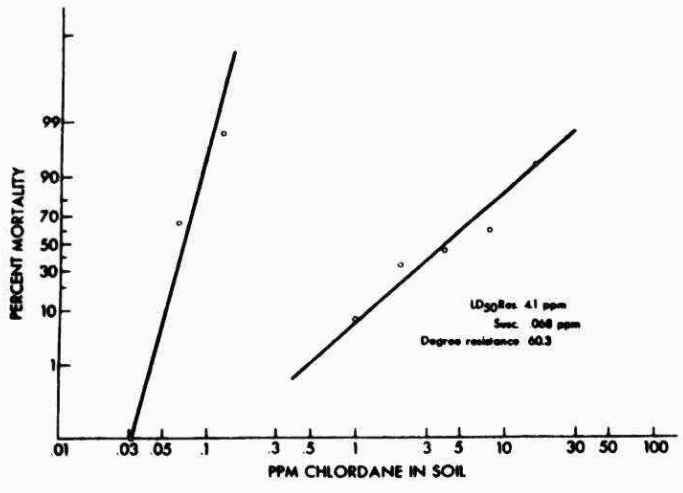

Figure 1.--Dose-mortality differences in susceptible and chlordane-resistant first instar Japanese beetle larvae held in chlordane incorporated sandy loam at 25 C ambient.

being resistant to the chlorinated hydrocarbon insecticides.

When mature eggs were available from each lot of beetles, the eggs were placed in chlordane-treated and untreated soil for incubation at 25 C. Eggs from each location were placed in seven containers of chlordane-treated soil at 4 ppm and three containers of untreated soil. As shown in Table 3, this dose of 4 ppm was more than 10 times the dose required to produce 100 per cent mortality of susceptible grubs. As soon as the earliest first instar larvae had transformed to seconds, the soil from all of the containers at a given location was screened for the live larvae. Table 4 indicates that a definite resistance to chlordane was present in beetles collected within a 2-mile radius of the golf course. The presence of resistance in three other sites, one within 2-4 miles and two within 6-8 miles of the golf course, is questionable due to the low numbers of larvae obtained in the untreated soils from these areas. However, since at least one normal larva was present in the treated soils,

Table 4.—Geographic distribution of chlordane-resistant Japanese beetles in Onondaga County, New York. 1972.

Radius from known site of resistant strain (miles)	No. samples beetles obtained from each radius	No. samples with susceptible or resistant 1st instar larvae[a]	
		Susceptible	Resistant
0	1	0	1
0-2	6	1	5
2-4	4	3	1 ?
4-6	4	4	0
6-8	3	1	2 ?
8-12	4	4	0
40+	3	3	0

a/ Seven containers of 30 eggs/treatment held at 25 C ambient in soil containing 4 ppm chlordane, counts made when second instar larvae first appeared in untreated soil. Absence of larvae indicated susceptible strain.

it appears likely that resistance is present beyond the 2-mile radius. Figure 2 shows the area where definite resistance occurs as well as the questionable areas. None of the samples taken beyond the 8-mile radius showed any evidence of resistance.

Field Control of Resistant Strains—Immediately upon determining the presence of the resistant strain in laboratory studies, field plots were established on infested turf at the golf course. Five materials were applied to 20 x 20 foot plots with five replications in a randomized complete block design. Treatments were made on April 28 during a cool (60's) cloudy day when the soil temperature at the 1-2 inch depth was 60 F.

Examinations for presence of larvae were made on May 26 by making five individual square foot diggings to a depth 4-6 inches in each plot. Samples were

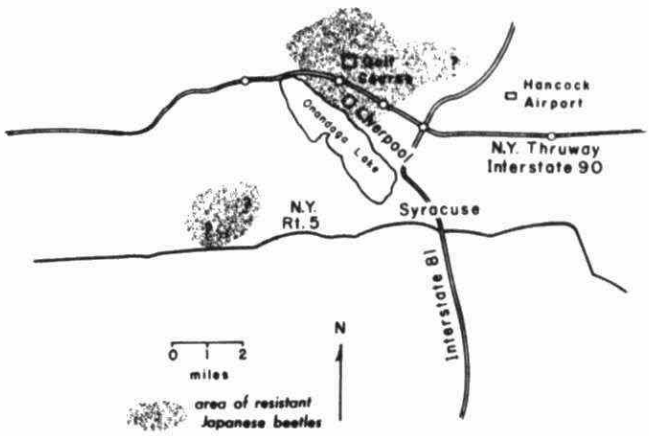

Figure 2.—Geographic location of chlordane-resistant Japanese beetle population in New York.

Table 5.—Control of chlordane-resistant second and third instar Japanese beetle larvae. Applications made April 28, 1972; surveyed May 26 and June 13, 1972. Liverpool, New York[a].

Insecticide and formulation		Lb. AI /acre[d]	Avg no. larvae/5 1 ft.^{2} surveys/plot[b]		% control[c]	
			5-26	6-13	5-26	6-13
Diazinon	2G	6	2.6 a	0.8 a	97	98
Carbaryl	5G	12	15.8 a	8.8 a	81	83
Chlorpyrifos	22% EC	2	25.4 b	11.0 a	70	78
Chlorpyrifos	0.5G	2	31.8 b	14.2 a	62	72
Pirimiphos-ethyl	2.1G	2	38.0 bc	11.6 a	54	77
Chlordane	10G	4	64.8 bc	40.2 b	22	20
Untreated		0	83.2 c	50.2 b	—	—

a/ Plots 20 x 20 feet, five replications/treatment.
b/ Means of each column followed by the same letter are not significantly different at the 1 per cent level according to Duncan's multiple range test.
c/ Based on Abbott's formula.
d/ Lbs/acre closely equivalent to ppm in laboratory tests on the basis of a 3-inch acre; $\frac{PPM}{1.0421}$ = Lb/acre; Lb/acre x 1.0421 = PPM.

taken inside a 3-foot border of each plot. Since there were numerous moribund larvae as a result of insecticidal poisoning, identical surveys were made again on June 13. The results shown in Table 5 indicate that chlordane showed no significant difference from the untreated checks. The degree of control was so low that evidence of resistance was definitely shown. All other compounds produced from moderate to acceptable levels of control with diazinon providing the highest level of control. Because of the large variations of larval recovery between plots of some of the treatments, large differences were required for significance.

CONCLUSIONS

On the results of the laboratory studies of first and third instar larvae and of field plots, the occurrence of a chlordane-resistant Japanese beetle population was clearly established. It is quite likely that there is considerable cross-resistance to the other chlorinated hydrocarbon compounds.

The occurrence of resistance in this particular area is not surprising since dieldrin and chlordane have been distributed in this area during the late 1950's and early 1960's primarily for control of the European chafer. The discovery of chlorinated hydrocarbon-resistant Japanese beetles makes it the second species in the family Scarabaeidae following the European chafer known to have developed resistance.

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Personnel at the New York State Agricultural Experiment Station whose services and assistance have been valuable include F. L. Consolie, who assisted in all phases of these studies; Gertrude Catlin, who collected adult beetles in Waterloo; and Rose McMillen, who prepared the illustrations.

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1/ Professor of Entomology, New York State Agricultural Experiment Station, Geneva, New York 14456

2/ Cooperative Extension Agent, Cooperative Extension Association of Onondaga County, Syracuse, New York 13202

3/ Concurrent to our discovery of this resistant strain, H. D. Niemczyk of the Ohio Agricultural Research and Development Center and K. O. Lawrence of the Japanese Beetle Laboratory, U. S. Department of Agriculture at Wooster, Ohio discovered and investigated a chlorinated hydrocarbon-resistant strain of the Japanese beetle in Ohio (personal communication).

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Japanese Beetle[1]: Evidence of Resistance to Cyclodiene Insecticides in Larvae and Adults in Ohio[2,3]

H. D. NIEMCZYK[4] and K. O. LAWRENCE[5]

Chlordane, aldrin, dieldrin, and heptachlor have been used extensively to protect lawns, golf courses, and other types of turf in Ohio from injury by Japanese beetle, *Popillia japonica* Newman. Depending upon the rate, protection for 3 or more years could be obtained with these insecticides. Chlordane was used successfully to control Japanese beetle grubs in the fairways of the golf course at The College of Wooster (CW), Wooster, Ohio, for about 14 years. In 1970, infestations of grubs were noted in the fairways in spite of the applications. Beginning in the spring of 1970, the applications of chlordane (5–8 lb AI/acre) were increased to 1 each spring and fall in an effort to control the grubs. However, samples taken May 25, 1972, from various fairways of the golf course showed that a larval population of 10–20/ft² existed. The study reported here was conducted to determine whether the poor control achieved could be attributed to development of resistance to chlordane in the population.

METHODS AND MATERIALS.—*Larval Test.*—On May 25, 1972, samples of 3rd-stage Japanese beetle larvae were obtained from the top 2 in. of soil in a treated fairway of CW, and from a turf area at the Ohio Agricultural Research and Development Center (OARDC), Wooster, which had not preivously been treated with cyclodiene insecticides. GLC analysis of samples from the top 2 in. of soil at CW taken at this time showed 2.10 ppm chlordane. These locations were about 2 miles apart.

[1] Coleoptera: Scarabaeidae.

[2] Approved by the Associate Director of the Ohio Agricultural Research and Development Center as Journal Article no. 79-72. Received for publication Aug. 14, 1972.

[3] The authors thank Dr. A. C. Waldron, Department of Entomology, Ohio State University, Columbus, for performing the GLC analysis, and the Upjohn Co., Kalamazoo, Mich., for assistance and cooperation. This paper reports the results of research only. Mention of a pesticide in this paper does not constitute a recommendation by the USDA.

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Table 1.—Results of laboratory tests for evidence of resistance to chlordane in 3rd-stage Japanese beetle larvae from Wooster, Ohio. 1972.

Days after treatment	Avg corrected % moribund (M) and dead[a] (D) larvae or pupae from 2 locations[b] at indicated equivalent rates of chlordane							
	4 lb AI/acre[c]				8 lb AI/acre[c]			
	OARDC		CW		OARDC		CW	
	M	D	M	D	M	D	M	D
4	0	0	0	0	0	0	0	4
7	84	4	0	0	84	8	0	4
10	76	8	0	0	84	12	4	4
15	80	20	0	0	56	44	4	4
18	60	36	0	0	56	44	0	4
21	60	36	0	0	28	72	0	4
25	60	40	0	0	16	84	0	4
28	44	56	0	0	12	88	0	8
% of test larvae which developed into adults	0		100		0		76	

[a] Mortality figures corrected for check mortality with Abbott's formula and based on 25 larvae, reared individually, from each location.

[b] Turf area at Ohio Agricultural Research and Development Center (OARDC) and fairway in the College of Wooster Golf Course (CW).

[c] Soil fortified with 10% chlordane dust to produce equivalent rates of 4 and 8 lb AI/acre.

Table 2.—Toxicity of 6 insecticides to adult Japanese beetle from 2 locations at Wooster, Ohio. 1972.

	48-hr avg corrected* % mortality at indicated % insecticide concentration									
	CW					OARDC				
Insecticide	0.0001	0.001	0.01	0.1	1.0	0.0001	0.001	0.01	0.1	1.0
Aldrin	11	0	18	18	29	3	0	37	100	100
Heptachlor	0	0	0	0	4	3	3	13	100	100
Chlordane	4	7	0	7	18	0	0	7	33	100
Dieldrin	7	0	7	4	0	0	3	7	100	100
Carbaryl	0	0	7	75	100	3	3	33	97	100
Diazinon	0	0	0	7	71	0	3	0	7	77

* Mortality figures corrected for check mortality by Abbott's formula and are the mean of 3 replicates of 10 beetles (5 ♂, 5 ♀).

Moist silt loam soil, which by GLC analysis contained an average of 1.785 ppm chlorinated hydrocarbon insecticides, was fortified with 10% chlordane dust to produce the equivalent of 4 and 8 lb AI/acre and was placed in covered 2×2-in. metal containers. Using 1 larva/container, 25 larvae each from OARDC and CW were caged at each rate. The check consisted of 25 larvae from each location caged in unfortified soil from the same source. A combination of redtop grass, rye grass, and white dutch clover seed (1:1:1 wt/wt) was mixed with all soil used, thus producing seedlings on which the caged larvae could feed. The containers of test insects were held in a laboratory maintained at 21±1.5°C.

The condition of the test insects was determined at 3 to 4 day intervals by emptying the contents of each container, adding seed and water if needed, and returning the soil and insects to the containers. Corrections for check mortality (4%) were made using Abbott's formula (Abbott 1925). The test for resistance was terminated as soon as the 1st adult was noted (28 days). However, observations continued until all test insects had either died or developed into adults.

Adult Test.—Adult beetles from OARDC and CW were collected July 17, 1972, using traps baited with phenethyl propionate + eugenol (7:3 vol/vol) (McGovern et al. 1970). The beetles were separated, caged according to sex and location, fed fresh sassafrass leaves, and conditioned at 26±1.5°C and 40±7% RH for 18 hr prior to treatment on July 19. Stock solutions (1%) of the test insecticides were prepared from analytical or technical-grade materials (98% carbaryl, 99% diazinon, 60% chlordane, 90% aldrin, 85% dieldrin, 99% heptachlor) using a mixture of acetone and olive oil, 19:1 (vol/vol) as the solvent. Three replicates of 10 beetles (5 ♂, 5 ♀), anesthetized with CO_2, were placed ventral side up on filter paper in 9-cm glass petri dishes. Each replicate was sprayed with 5 ml of each insecticide in concentrations of 0.0001%, 0.001%, 0.01%, 0.1%, and 1.0%. The checks consisted of 2 sets of 3 replicates of 10 beetles (5 ♂ and 5 ♀) from each location, the 1st consisting of a random sample of unsprayed anesthetized beetles taken near the end of the test, and the 2nd consisting of beetles sprayed with the solvent. The solutions were applied using a Potter Spray Tower generally modified as described by Harris and Mazurek (1964). Fifteen sec were allowed for delivery and 15 sec for settling. The group of 10 treated beetles was removed from the petri dish and placed in a paper cup containing a slice of ripe apple. The cup was covered with a metal screen and placed in the same room in which the insects were conditioned prior to treatment. The number of dead and moribund beetles was recorded 24 and 48 hr after treatment. Corrections for check mortality (3.3% for both types of checks from both locations) were made using Abbott's formula (Abbott 1925).

RESULTS AND DISCUSSION.—The results of the larval and adult tests (Tables 1 and 2, respectively) clearly show a high level of resistance to chlordane. The adult beetles from CW were also highly cross-resistant to aldrin, dieldrin, and heptachlor. Diazinon and carbaryl were equitoxic to adults from both locations. An examination of the adult data taken 24 hr after treatment showed: (1) both sexes from both locations were equisusceptible to diazinon and carbaryl: (2) both sexes from CW were equisusceptible; (3) males from OARDC were nearly twice as susceptible as females to the cyclodiene insecticides.

Our studies were conducted concurrently with investigations being conducted during the spring and summer of 1972 by Dr. H. Tashiro, Geneva, N. Y. on a chlordane-resistant strain of Japanese beetle near Syracuse, N. Y.

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European Chafer:[1] Noncyclodiene Insecticides for Control of Grubs[2,3]

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ABSTRACT

In studies to investigate the effectiveness of new insecticides, Bay 37289 (*O*-ethyl *O*-(2,4,5-trichlorophenyl) ethylphosphonothioate) gave complete control of larvae of *Amphimallon majalis* (Razoumowsky) when 5 pounds per acre were either incorporated in the soil or applied as a surface treatment. Also, assays of soil samples from field plots showed that incorporated compounds used at 10 or 20 lb/acre would give 2 or 3 years of control, respectively, in established turf. In addition, Bay 37289 controlled the Japanese beetle, *Popillia japonica* Newman, in a mixed-field population of Japanese beetles and European chafers almost as well as the chafers. Other materials tested, diazinon (slow-release and regular formulations), Dyfonate® (*O*-ethyl *S*-phenyl ethylphosphonodithioate), American Cyanamid CL-47031 (cyclic ethylene *P,P*-diethyl phosphonodithioimidocarbonate), and propoxur were inferior to Bay 37289.

The residual activity of the cyclodiene insecticides dieldrin and aldrin was a great advantage in controlling larvae of the European chafer, *Amphimallon majalis* (Razoumowsky), and the Japanese beetle, *Popillia japonica* Newman. Applications could be made at any time when the ground was not frozen, and a single application provided a high degree of control for many years (Gambrell et al. 1968). Therefore, in view of the restrictions on use of these insecticides and the possibility that the insects could become resistant, we investigated insecticides that could be substituted, with the hope of detecting materials that would give a single season's control after either surface application to sod or incorporation into the soil.

Experiments and Results.—*Laboratory Screening Tests.*—A large number of candidate insecticides were screened at the European Chafer Investigations Laboratory at Geneva, N. Y., from 1964 to 1967 by incubating field-collected 3rd-stage larval European chafers in trays containing 20 lb of toxicant/3-in.-acre of Honeoye sandy loam soil (4 trays of 25 larvae/mixture). The trays were held at 21°C, seeded with redtop grass (to provide food for the larvae), and watered as needed. Larvae were examined weekly for 6 weeks, and dead larvae were noted and removed. Mixtures causing 100% mortality were tested for another 6 weeks with 25 additional larvae, and those retaining this level of toxicity (American Cy-

[1] Coleoptera: Scarabaeidae.
[2] This paper contains the results of research only. Mention of a pesticide or a proprietary product does not constitute recommendation or endorsement by the USDA.
[3] Approved by the Director of the New York State Agricultural Experiment Station as Journal Paper no. 1806, dated May 22, 1970.
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Table 1.—Percent control[a] of 1st-stage larvae of the European chafer in composite soil samples taken at increasing intervals from field plots treated in 1966–67 with the indicated insecticides.

Materials	Rate of application (lb/acre)	Months after incorporation (April 1966) at indicated rate				Months after treatment (May 1967) at indicated rate			
						Incorporation		Surface treatment	
		3.25	15	27	45	2.5	15	2.5	15
Diazinon 5G (slow-release)	1.25					14.7		17.2	
	2.5	20.0	23.9	0		45.5	0	23.7	0
	5	71.6	12.8	0		76.3	0	14.7	0
	10	100	24.9	1.4		100	0	62.9	0
	20	100	33.3	0					
Dyfonate 5G	1.25			10.3		22.6		23.7	
	2.5	100	0	17.7		95.4	13.2	32.6	13.3
	5	100	5.7	19.3		99.3	5.0	70.5	16.3
	10	100	8.2	22.2					
	20	100	7.6	22.2					
Bay 37289 10G	1.25					66.6		25.6	
	2.5	100	10.4	18.6	0	100	6.6	100	7.4
	5	100	16.6	31.0	0	100	52.9	100	43.1
	10	100	100	14.9	0				
	20	100	100	100	44.1				
Dieldrin	1.25					100	100	100	100
	2.5	100	100	100	100	100	100	100	100

[a] Corrected by Abbott's Formula. Percentage survival in controls = 44–78%.

anamid CL-47031 (cylic ethylene *P,P*-diethyl phosphonodithioimidocarbonate), Bay 37289 (*O*-ethyl *O*-(2,4,5-trichlorophenyl) ethylphosphonothioate), propoxur, diazinon (regular and slow-release formulations), and Dyfonate® (*O*-ethyl *S*-phenyl ethylphosphonodithioate)) were studied further.

Toxicity of Residues 1st-Stage Larvae.—To determine the continuing toxicity of the candidate materials to 1st-stage larval European chafers, we tested the compounds as granular formulations (Table 1) by incorporating them in the soil during April 1966 in twice-replicated, randomized 1/100-acre plots at Geneva, N. Y.; dieldrin was used as the standard. The insecticides were applied with a precalibrated spreader to a field that had been previously cultivated, and they were worked 3 in. into the soil with a rototiller. Plots were not watered during the study. Then at 3.25, 15, 27 and 45 months past treatment, we removed fifty 1⅝×3-in. soil cores from each plot and returned them to the laboratory where they were composited and screened to remove debris. The soil was then distributed into 4 or 8 clay pots, of 6½-in. diam, and 25 or 30 chafer eggs were introduced into each pot and covered with about 1 in. of soil. The pots were seeded with redtop and blue grass, held indoors in moist sphagnum moss at 20°C, and watered as needed. After 90% or more of the grubs in pots containing untreated soil had transformed to the 2nd stage, we examined the soil in pots containing treated soil and recorded the numbers of larvae present.

Again, in May 1967, we established a 2nd set of plots and treated them (both surface application and incorporation in soil) with the materials shown to be most effective in the 1966 tests. The 1967 plots were sampled at 2.5 and 15 months posttreatment.

Table 1 presents the results of both tests. We obtained 14.3, 90.1, and 97.5% control from 20 lb/acre of propoxur, CL-17031, and diazinon (regular formulation), respectively, in the 1st test at 3.25 months, so we discontinued sampling these plots and have not included the results. Diazinon (slow-release formulation) at 10 lb/acre, Dyfonate at 5 lb/acre, and Bay 37289 at 2.5 lb/acre gave 99–100% control at 2.5–3.25 months past incorporation. Bay 37289 incorporated at 10 and 20 lb/acre gave 100% control for 15 and 27 months, respectively, and complete control at 2.5 months after surface application of 2.5 lb/acre. Diazinon (slow-release) applied at 10 lb/acre, Dyfonate at 5 lb/acre, and Bay 37289 at 2.5 lb/acre gave 62.9, 70.5, and 100% control, respectively, 2.5 months after application to the surface.

Initial Amounts Needed to Kill 1st-Stage Larvae.—To determine the amounts of Bay 37289, diazinon, Dyfonate, and dieldrin (control) required to kill all incubated 1st-stage larvae and to determine the actual amounts of insecticide remaining after 27 months in the plots treated in April 1966, we subjected soil from these plots to chemical analysis in 1968.

For the chemical analyses, aliquots from the 27-month composite samples were removed after screening, slurried with water, and extracted with hexane. Required cleanup was minimal, and all compounds were completely recovered during extraction. Also, by this procedure, we avoided extensive decomposition of Bay 37289 during air-drying of the soil before analysis (Anonymous 1965). A tritium detector was used to identify dieldrin and Bay 37289, and a flame photometric detector with a phosphorus filter was used to detect diazinon and Dyfonate. The gas-liquid chromatographic operating parameters were ⅛-in. glass columns maintained at 200°C. Chlorinated compounds were resolved on 5% DC 11® on 60/80 mesh Chromasorb® and phosphate on 2% Reoplex® on 80/10-mesh Gaschrom Q®. Peak areas for each material were standardized against the pure reference compounds.

Table 2.—Residues of insecticides found by chemical analysis 27 months after incorporation in soil and the amounts needed in freshly prepared soil mixtures for 100% control of 1st-stage larvae of European chafers.

Compound	Insecticide (lb/acre)		
	Originally applied to field plots	Remaining in plots after 27 months	Required for 100% control
Bay 37289 10G	2.5	0.05	0.8
	5	.1	
	10	.2	
	20	1.0	
Diazinon 5G	2.5	.03	.8
	5	.04	
	10	.06	
	20	.09	
Dyfonate 5G	2.5	.01	.8
	5	.02	
	10	.06	
	20	.09	
Dieldrin	2.5	1.5	.6

Also, we prepared fresh soil-insecticide mixtures containing 0.2, 0.4, 0.6, 0.8, 1.0, 1.4, and 1.8 lb/3 in.-acre; then 2 days later, 30 embryonated eggs were incubated in ten 6½-in.-diam pots of each mixture and incubated indoors at 28°C. The pots were seeded and watered, and toxicity to the larvae was determined as previously indicated.

Table 2 presents the results of the chemical analyses and the bioassays. Dyfonate, diazinon, and Bay 37289 all gave 100% control when they were applied at 0.8 lb/acre, and dieldrin gave equal control when it was applied at 0.6 lb/acre. The chemical analyses showed that soil treated with Bay 37289 (originally applied at 20 lb/acre) and dieldrin (applied at 2.5 lb/acre) soil contained more insecticide than the least amount required to kill all larvae in freshly prepared soil-insecticide mixtures. In contrast, the amount of diazinon and Dyfonate required to kill 100% of the larvae (0.8 lb/acre) was not detected in any field samples, and, accordingly, complete mortality (Table 1) did not occur.

Control of European Chafer Larvae in the Field.—In 1969, we made 2 studies to determine the amounts of Bay 37289 and Dyfonate required to control newly hatching European chafer larvae in the field. In the 1st study, Bay 37289 and Dyfonate were applied to 20×20-ft turf plots in Webster, N. Y., at 2.5, 5, 10, and 20 lb/acre during June, a time when the turf contained a mixed population of European chafers and Japanese beetles. Each material was applied in a separate 5×5 Latin square with a Scott® spreader as described by Tashiro and Fiori (1969). The plots were watered immediately after application to wash the insecticide from the vegetation. Four months later, 10-ft samples 3–4 in. deep were taken from each plot, and the numbers of larvae were recorded. Since a mixed population of European chafers and Japanese beetles was present, we recorded the numbers of both species surviving.

For the 2nd test, the soil in 12×12-in. clay pots was treated with Bay 37289, applied to the surface or incorporated, at 2.5, 5, 10, or 20 lb/acre at Geneva. Treatments were replicated 3 times. Pots receiving surface treatments had had turf established prior to treatment, and those receiving incorporated treatment were seeded immediately after treatment. All treatments were replicated 3 times. Then 6 weeks after the insecticide was applied, 10 ♀ and 20 ♂ adult chafers were caged over the pots with conical steel cages. The numbers of surviving larvae per pot were determined 4 months after the adults were introduced.

In the turf plots at Webster (Table 3), Bay 37289 gave complete control of European chafer larvae 4 months after application of 5, 10 or 20 lb/acre, but complete elimination of Japanese beetle larvae required 10 lb/acre. In contrast, 20 lb/acre of Dyfonate was necessary for 100% control of chafer larvae, but this amount reduced Japanese beetle larvae only by about 92%.

In the test at Geneva, the European chafer larvae were completely controlled in all treatments with Bay 37289. The controls returned an average of twenty-seven 3rd-stage larvae/pot.

DISCUSSION.—Although our tests showed that dieldrin is the most effective soil insecticide against European chafer larvae, Bay 37289 appears promising as a substitute. In the several field and laboratory tests we carried out, Bay 37289, when applied at a rate of 5 lb/acre between April and early June, gave complete control for 1 year, and 2.5 lb/acre applied during the same period killed about 95% of the subsequent larval brood. When Bay 37289 was incorporated in the soil at 10 or 20 lb/acre, complete control of incubated larvae was still being obtained after 15 and 27 months, respectively. However, the results of this test (Table 1) also indicated that residues in plots originally containing 5, 10, or 20 lb/acre of May 37289 began to lose their effectiveness at 12, 27 and 45 months, respectively, after application. Studies are being continued to determine the periods that control can be expected from the surface applications of Bay 37289 to established turf.

The overall performance of Dyfonate, diazinon (slow-release and regular formulations), American Cyanamid CL-47031, and propoxur was inferior to

Table 3.—Percent control[a] of European chafers and Japanese beetle larvae in field plots in Webster, N. Y., 4 months after surface applications of the indicated insecticides.

Material	Rate of application (lb AI/acre)	% control of	
		European chafers	Japanese beetles
Bay 37289 15G	2.5	94.8	83.2
	5	100	98.4
	10	100	100
	20	100	100
Dyfonate 10G	2.5	0	26.4
	5	57.4	13.8
	10	93.6	63.5
	20	100	91.8

[a] In the controls the average number of grubs/10²-ft sample per plot was 1.8 for the European chafer and 9.1 for the Japanese beetle. Percentages corrected by Abbott's formula.

that of Bay 37289. However, Dyfonate does appear promising for seasonal control, and the timing of application is not critical, but surface applications become ineffective rapidly (Tables 1, 3). Diazinon (slow-release and regular formulations) and Dyfonate may provide control for a single season as surface or incorporated treatments if they are applied in mid-July.

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1972, unpublished data). The most prominent predators, *Geocoris uliginosus* (Say) (Hemiptera: Lygaeidae) and *Lasiochilus sp.* (Hemiptera: Anthocoridae) appear to exert considerable pressure on chinch bug populations.

Control.—When southern chinch bug populations reach damaging proportions, applications of pesticides are the only reliable control tool at the moment. Kerr (1966b) gave details on effective insecticides and methods to be used by commercial operators, and Brogdon and Kerr (1964) gave directions for control by home gardeners. Several materials provide excellent control including: Akton® (*O*-(2-chloro-1-(2,5-dichlorophenyl) vinyl) *O,O*-diethyl phosphorothioate), carbophenothion, chlorpyrifos, diazinon, ethion, propoxur, and propyl thiopyrophosphate. These and several others were shown to be very effective against the southern chinch bug by Reinert (1972a).

Both granulated and spray formulations are effective, but most commercial lawn pest control is done by spraying. Drenching sprays of at least 15 gal/1000 ft² are essential to achieve control in thick, southern Florida turf thatches.

Hairy Chinch Bug

Bionomics.—Cruz and Streu (1973) have recently studied the bionomics and population dynamics of *B. l. hirtus*. Their findings in New Jersey are probably representative of much of that section of the Eastern Seaboard. Hairy chinch bugs feed on bentgrasses, bluegrasses, fescues, and have become serious problems on zoysiagrasses where the latter have been planted extensively.

These chinch bugs overwinter as adults in protected places like tufts of grass, under plant debris, and in dense thatch. Hibernation did not end until the last week in May in the New Jersey study. There are 2 generations a year, with peak populations in September. The time from egg to adult was estimated at about 43 days in the field. The mean number of eggs laid was 54/female with 1.56/day.

Cruz and Streu collected a large predator fauna in turfgrass, and found 8 species of arthropods which fed on early stages of *B. l. hirtus* in the laboratory. Studies so far indicate that predatory mites and *Geocoris bullatus* (Say) (Hemiptera: Lygaeidae) are of importance in regulating hairy chinch bug populations.

Control.—Many insecticides can provide excellent control of the hairy chinch bug. Streu and Cruz (1972) achieved season long control with 1 application of chlorpyrifos. Studies in New Jersey and elsewhere have shown excellent control with carbophenothion, ethion, carbaryl, propylthiopyrophosphate and others (Schread 1970, Polivka 1963, Streu 1969).

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BIONOMICS AND CONTROL OF ROOT FEEDING INSECT PESTS: GRUBS AND BILLBUGS

by Haruo Tashiro

White grubs and allies (Scarabaeidae) (Ritcher 1966) and species of *Sphenophorus* and *Hyperodes* (Curculionidae) are the most destructive below-ground turf insects. Scarabaeid grubs are general feeders of fibrous roots; adults are generally voracious foliage feeders. Billbug larvae destroy roots of grass plants, while adults are generally voracious foliage feeders. Billbug larvae destroy roots of grass plants, while adults feed and oviposit in the stems of the same plants.

Similarities in the damage caused by these two groups are superficial, as both cause turf to die through root destruction. Similarities end here. Most scarabaeid grubs damage lawns during spring and fall leaving the surface soft and spongy as the fibrous roots are consumed. Severely damaged turf, even though still green, can usually be rolled back like a rug. Billbug damage occurs during mid-summer as the larger larvae leave the grass stems to feed on the crowns and roots. A spongy surface does not occur and the turf cannot be easily peeled.

Scarabaeid Grubs

Native Grubs.—The most numerous and widespread of turf pests, *Phyllophaga* spp., occur throughout the U.S. and Canada but most commonly in the north central states to the eastern seaboard (Luginbill and Painter 1953). Of some 150 species, *P. anxia* (LeConte), the most widespread, is recorded from all but 5 of the 48 contiguous states. A 3-year life cycle is the most common among *Phyllophaga* spp. Individuals spend 2 winters as a grub; the first as a 2nd stage larva and the second as a 3rd stage larva. A pre-emerged adult hibernates the third winter and emerges the following spring to feed and lay eggs (Hammond 1940).

Important native white grubs with 1 year life cycles

are the northern masked chafer, *Cyclocephala borealis* Arrow, the southern masked chafer, *C. immaculata* (Olivier), and the green June beetle, *Cotinis nitida* (Linnaeus). The grub of the latter has an unusual habit of crawling central side up.

Introduced Grubs.—Important introduced grubs include: the Japanese beetle, *Popillia japonica* Newman, the European chafer, *Amphimallon majalis* Razoumowsky; the oriental beetle, *Anomala orientalis* Waterhouse; and the Asiatic garden beetle, *Maladera castanea* Arrow.

Bionomics of the Japanese Beetle.—The wide distribution, host range and damage caused by grubs and adults make the Japanese beetle one of the most destructive insects in the U.S. (Fleming 1972). It occurs in Ontario, Canada and in every state east of the Mississippi River except Florida, Louisiana, Minnesota and Wisconsin. (USDA Japanese Beetle Quarantines, Revised Jan. 24, 1972). It overwinters predominantly as 3rd stage grubs below the frost line, resumes feeding near the soil surface during spring, and pupates in late spring and early summer. Beetles fly during warm sunny days mainly in July and August, when they feed and mate on host plants before returning to the soil in the evening to oviposit.

The last range of the beetle includes some 300 plants with grapes, roses, peaches, apples, and maples preferred. By fall the grubs are predominantly 3rd stage and feed vigorously until cold weather forces them into hibernation. Turf damage is greatest during fall and spring when the large grubs feed just below the soil surface.

During a series of dry summers, eggs desiccate resulting in diminished larval populations. A series of wet summers produce a resurgence of high populations through greater egg survival. Populations of the Japanese beetle and the European chafer have increased during the past 2-3 years for the same reason.

Natural Enemies.—Several biological agents reduce Japanese beetle populations. Milky disease caused by *Bacillus popilliae* Dutky and *B. lentimorbus* Dutky are very effective pathogens of Japanese beetles in the more southern areas. Various strains of either bacteria such as *anxia, hirticula, Cyclocephala* and *Amphimallon* infect other species of grubs as the names imply. These are ideal insect pathogens since the resistant spores lie dormant in the soil until ingested by host grubs. Once an epizootic develops the soil becomes heavily charged with spores and the effectiveness of the disease is heightened without further artificial inoculations.

Tiphia vernalis Rohwer and *T. popilliavora* Rohwer introduced from the Orient are highly effective hymenopterous parasites of the Japanese beetle grubs; native species of *Tiphia* parasitize native grubs.

Birds and mammals, namely skunks, moles and shrews feed on grubs. The mammals may cause more damage to the turf by their rooting and burrowing activities than the insects do.

Scarabaeid Grub Control.—The population levels of scarabaeid grubs that justify treatments is a difficult decision to make. In New York, we suggest treating when there are 2 or more grubs per square foot of sparse lawn or 4 or more of vigorous lawn. Figures range from 1 to 5 white grubs per square foot from various areas throughout the country. The chlorinated hydrocarbon insecticides made grub control an extremely efficient practice. A single application provided 10 to 15 years of protection. In recent years it was shown that dieldrin at 0.5 lb and chlordane at 4 lb per acre, a sixth and about half, respectively, of previous recommendations, controlled Japanese beetle and European chafer larval populations for at least 5 seasons (Unpublished data).

Chlorinated hydrocarbon resistant European chafers near Rochester, N.Y. (Tashiro *et al.* 1971) and the concurrent discovery of similar resistance in Japanese beetles during April 1972—in Liverpool, New York (Tashiro and Neuhauser 1973) and in Wooster, Ohio (Niemczyk and Lawrence 1973) plus legislative actions against these compounds complicates the control of these larvae. Where resistance occurs, diazinon and chlorpyrifos are the most effective available materials. These insecticides must be used annually and be critically-timed to coincide with presence of young grubs.

In Texas, *Phyllophaga crinita* Burmeister is developing resistance to the chlorinated hydrocarbons and diazinon is the preferred replacement compound (Frankie and Hamman 1972).

Billbugs

The two most troublesome species are the bluegrass billbug, *Sphenophorus parvulus* Gyllenhal, which attacks cool season grasses (namely bluegrass), and the hunting billbug, *S. venatus vestitus* Chittenden that attacks warm season grasses, namely, zoysia and bermuda. Both species are widespread. The bluegrass billbug is troublesome across the northern half of the nation while the hunting billbug is more common in the southern half and Hawaii. In New York, the former suddenly became a serious turf pest in the Rochester area in 1967 (Fiori *et al.* 1968) in the same area where the chlorinated hydrocarbon resistant European chafers occurred. This problem largely disappeared within 3 years.

Bionomics of Billbugs.—Adults overwinter in protected areas and emerge during the first warm days of spring. They feed and deposit eggs in stems just above the soil surface. Young larvae feed in the stems, but later stages migrate to the soil and feed on the roots, crown and rhizomes producing frass superficially resembling fine sawdust. Larval growth is completed during summer; they pupate in the soil and become adults before fall. Bluegrass billbug adults seem to wander aimlessly on pavements adjacent to lawns on warm sunny fall days, apparently in search of hibernation sites.

Billbug Control.—Diazinon, carbaryl and chlorpyrifos are effective compounds. In New York we are recommending a June to early July application when an average of one adult per minute is seen on adjacent paved surfaces.

Turfgrass Weevil

Hyperodes spp., are not a billbug, but their life cycle and damage caused are similar to that of the bluegrass billbug. Annual bluegrass on Long Island and adjacent areas is damaged; large patches of turf are killed in late May and early June (Cameron and Johnson 1971). Diazinon and chlorpyrifos are effective when applied in mid-April and again in mid-May.

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Toxicity of 6 insecticides to cyclodiene-susceptible (S) and cyclodiene-resistant (R) 3rd stage larvae of the Japanese beetle from Wooster, Ohio - 1974.

Insecticide	48-hr avg. corrected[a] % mortality at indicated % insecticide concentration							
	0.001		0.01		0.1		1.0	
	S	R	S	R	S	R	S	R
Chlordane	5	0	0	5	90	5	100	10
Dieldrin	0	0	95	0	100	0	100	5
Diazinon	0	0	0	0	100	100	100	100
Dasanit	0	0	100	80	100	100	100	100
Dursban	10	10	5	20	100	100	100	100
Carbaryl	11	0	100	100	100	100	100	100

a/ Mortality figures corrected for check mortality by Abbott's formula.

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Susceptibilities of European Chafer and Japanese Beetle Grubs to Chlordane and Dieldrin: Suggesting Reductions in Application Rates[1]

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ABSTRACT

Laboratory tests were conducted to compare the relative susceptibilities of the European chafer, *Amphimallon majalis* (Razoumowsky), and the Japanese beetle, *Popillia japonica* Newman, as 1st- and 3-instar larvae to chlordane and dieldrin. As 1st instars, it required >1.46 ppm chlordane and 0.84 ppm dieldrin to eliminate the chafer compared with 1.25 ppm chlordane and <0.21 ppm dieldrin to eliminate the Japanese beetle. Grubs were more susceptible at fluctuating temperatures than at a constant temperature. As 3rd instars, it required 1.6 times more chlordane and 2.3 times more dieldrin to produce comparable results in the European chafer as in the Japanese beetle.

Lawn plots harboring mixed populations of the 2 species were treated with 4 levels each of chlordane and dieldrin during June 1968. Fall surveys 4 months later showed that both species were equally susceptible to dieldrin. European chafer grubs were more susceptible to chlordane than Japanese beetle grubs. From the results of 1 season, it appears that rates of application for protecting lawns against grub damage can be lowered from the currently recommended 11.2 kg/ha of chlordane and 3.4 kg/ha of dieldrin (equivalent to 10 and 3 lb/acre, respectively) to 4.5 and 1.1 kg/ha rates (equivalent to 4 and 1 lb/acre) for both species.

In spite of the ever increasing restrictions being placed on the use of cyclodiene insecticides in agriculture, in all probability some of these compounds will continue in use for an indefinite future, particularly against non agricultural pests. As yet no equally effective substitutes for chlordane and dieldrin have been found for controlling scarabaeid grubs in turf. One method of partially decreasing the primary objection to these compounds, that of long persistence, could be accomplished through significant reduction in the initial rates of application.

The initial recommendations of 22.4 kg of chlordane and 5.6 kg of dieldrin/hectare (equivalent to 20 and 5 lb/acre) for control of the European chafer, *Amphimallon majalis* (Razoumowsky), were based partially on the knowledge that 3rd instar grubs of the chafer were more difficult to kill than those of the Japanese beetle, *Popillia japonica* Newman, and partially on the necessity for virtually eliminating grubs to satisfy quarantine regulations. Subsequently, rates were reduced to 11.2 and 3.4 kg (equivalent to 10 and 3 lb, respectively), to coincide with recommendations for the control of the Japanese beetle when tests indicated that the 1st-instar chafer grubs were nearly as susceptible as those of the Japanese beetle. However, data were obtained at 2 different laboratories (on the Japanese beetle at the Entomology Research Division's Japanese Beetle Laboratory, Moorestown, N. J., and on the European chafer at the New York State Agricultural Experiment Station, Geneva) where conditions at the 2 laboratories were known to be dis

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similar. Consequently, knowledge of the relative susceptibilities of the 2 species continued to be only partially resolved.

More recently, we have been able to compare the susceptibility of 1st- and 3rd-instar grubs of the 2 species to chlordane and dieldrin by simultaneously exposing grubs of both species to identical test procedures. Moreover, lawn plots containing mixed populations of the 2 species were treated with insecticides and have been sampled to obtain data on the relative susceptibilities of the grubs in the field. This paper summarizes data obtained in laboratory and field studies on the relative susceptibility of grubs of the 2 species, and suggests that lower rates may be used for lawn protection where quarantine regulations do not apply.

METHODS AND MATERIALS.—*Laboratory Tests.*—Toxicity tests on 1st- and 3rd-instar grubs were conducted in the laboratory by infesting insecticide-treated soils with mature eggs or grubs. Chlordane and dieldrin, 10% G, were thoroughly incorporated into moist friable Honeoye fine sandy loam that had previously been passed through 2.36 mesh/cm screen to remove debris. At the time of incorporating insecticides, seeds were also added to germinate and provide food for the grubs. Red top and bluegrass seeds were used for 1st-instar tests and domestic rye grass and sweetclover seeds for 3rd-instar tests. The number of survivors was determined by passing the soils through screens to recover the grubs.

First-Instar Tests.—Beetles of each species were held in friable soil to obtain eggs. Tests were conducted by introducing eggs into 14-cm clay pots containing insecticide-treated soil to about 2.5 cm from the top. Prior to filling a pot, a fine plastic screen was placed over the drain hole to prevent grubs from escaping. Insecticidal levels in soil included 7 concentrations, 0.21–1.46 ppm, chlordane (AI) and 5 concentrations, 0.21–1.04 ppm, dieldrin (AI) with 0.21 ppm increments of each insecticide. At each concentration and with each species, 30 eggs were introduced into each of 14 pots; 7 of these were held indoors at a constant soil temperature of 21°C, and 7 were held outdoors in a coldframe where they were subjected to daily fluctuations in temperature and other weather conditions. The average outdoor soil temperature was ca. 18°C (range 14.4–25.6°C). With the pots placed in saturated peat both indoors and outdoors, very little watering was necessary to prevent drying of the soil. When 90+% of the grubs in the untreated controls had transformed to 2nd instars (about 6 weeks after introducing eggs) pots were examined for grub survival.

Third-Instar Tests.—Field-collected 3rd-instar grubs of the 2 species were introduced into soils treated at concentrations of 1.0, 2.1, 4.2, and 8.4 ppm chlordane (AI) and 0.5, 1.0, 2.1, and 4.2 ppm dieldrin (AI). Forty grubs of each species were introduced into each of 4 wooden trays (27×27×7.5 cm ID) nearly filled with soil giving four 40-grub replicates/treatment. Grubs were placed on the soil surface and permitted to burrow into the soil on their own accord. All trays were held indoors at a constant soil temperature of 21°C. Examinations were made at weekly intervals for 5 weeks by screening the soils to recover the grubs and returning them for further exposure in the same soil.

Field Tests. Lawn plots established on Eel silt loam at Webster Park, Webster, N.Y. June 17, 1968, in an area known to have supported moderately high populations of European chafer grubs for at least 6 seasons and relatively low but increasing populations of Japanese beetle grubs. Applications of 10% G were made on the lawn with each insecticide. Chlordane was applied at rates of 0, 1.1, 2.2, 4.5, and 9.0 kg AI/ha and dieldrin at rates of 0, 0.6, 1.1, 2.2, and 3.4 kg AI/ha. Each insecticide was distributed in 6.1×6.1-m plots arranged in 5×5 Latin square design. A 0.6-m Scott's spreader (Model no. 75-3, O. M. Scott & Sons, Marysville, Ohio) was used to spread the insecticides over the entire plot once. The entire volume of each preweighed amount could not be delivered through the spreader. Therefore, residues were permitted to fall into a trough attached to the bottom of the hopper and were then transferred to a large salt shaker. The amount in the shaker was then distributed over the entire plot to give as even a distribution as possible. After application each plot was watered to wash the insecticides off the vegetation.

Grub populations present in each plot were determined Oct. 1 (± 1 day) 4 months after application. Ten diggings of 0.1 m^2 were made in each plot to a depth of ca. 15 cm. Diggings were evenly spaced on a diagonal from the SW to the NE corner of each plot (Fig. 1). Precise and uniform positioning of these diggings will permit surveys in subsequent years without sampling areas previously dug. Grubs from each digging were placed in a shallow tray containing a little soil to reduce cannabalism and were identified in the field by their rastral patterns (Fig. 2) using a 10× magnification of a stereomicroscope.

RESULTS.—*First-Instar Tests.*—First-instar European chafer grubs were more difficult to kill than Japanese beetle grubs (Table 1). Elimination of chafer grubs was not achieved with 1.46 ppm indoors (the highest level used) but was achieved with 1.25 ppm outdoors, while Japanese beetle grubs were eliminated with 1.25 ppm indoors and with 0.63 ppm outdoors. Chafer grubs were eliminated with 0.84 ppm of dieldrin indoors and with 0.21 ppm outdoors, while Japanese beetle grubs were eliminated with 0.21 ppm (the lowest concentration) under both conditions. Grubs were slightly more susceptible at fluctuating temperatures than at a constant temperature, even though the average temperature outdoors was lower than the constant indoor temperature. Fleming et al. (1962) showed that the estimated requirements to eliminate newly hatched Japanese beetle grubs varied from 0.13

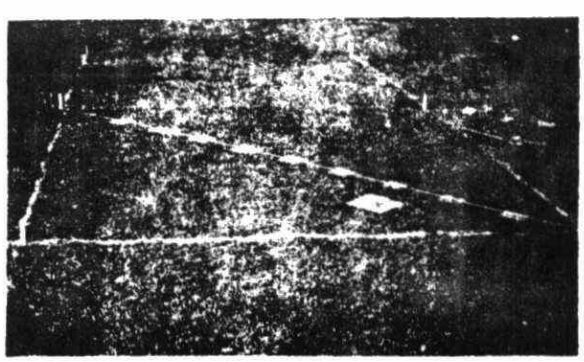

FIG. 1—Method of sampling in each plot. Soil cut to a depth of 15 cm around each 0.1 m^2 board. Precise locations of sampling units will permit sampling undisturbed turf for several years.

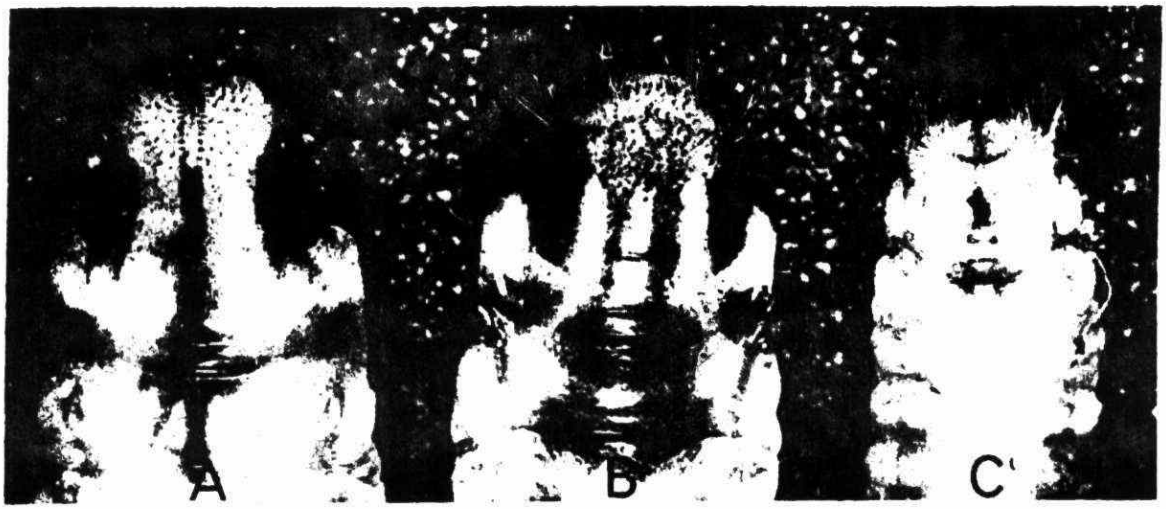

FIG. 2.—Rasters used as identifying characters of larvae. A. European chafer, B. Japanese beetle, and C. Asiatic garden beetle.

to 2.21 lb of chlordane and 0.11 to 1.26 lb of dieldrin/3-in. acre depending on the 9 or 10 sandy to silt loam soil types and series involved. This fact emphasizes one of the complexities in soil-insecticide relationships.

Third-Instar Tests.—Toxicity of chlordane and dieldrin to 3rd-instar grubs showed that the European chafer grubs were more difficult to kill than Japanese beetle grubs (Table 2). With both insecticides, mortality of chafer grubs was lower than that of Japanese beetle grubs in 17 out of 20 possible comparisons. At insecticide levels and exposure periods where direct comparisons can be made, it required up to 2× more chlordane and up to 4× more dieldrin to produce the same results in chafer as it did in Japanese beetle grubs.

In a more precise comparison, by graphing mortality percentages against insecticide levels on log probability scales for the 4-week interval, estimates of LD_{50} concentrations of chlordane were 2.15 ppm for chafer grubs and 1.33 ppm for Japanese beetle grubs, a ratio of 1.62:1. The LD_{50} concentrations of dieldrin were 2.40 ppm for the chafer grubs and 1.04 ppm for Japanese beetle grubs, a ratio of 2.32:1.

Field Tests.—Surveys made 4 months after application revealed the presence of mostly 3rd-instar grubs of both species but reflected the survivals following insecticidal activity on 1st-instar grubs. Judged from surveys made on the previous generation of grubs, the native populations were expected to be roughly comparable for the 2 species. In addition, a light population of the Asiatic garden beetle, *Maladera castanea* (Arrow), and a scattering of native white grubs, *Phyllophaga* spp., were expected. However, from the density of European chafer beetle flights which were seen during early July in the immediate vicinity of the plots, and the lack of Japanese beetle adults during early August or evidence of their feeding on surrounding vegetation, we expected a heavier population of chafer grubs than Japanese beetle grubs. It was thus a surprise to find that there was about 5× more Japanese beetle grubs than chafer grubs in the untreated plots. Results with chlordane indicated that chafer grubs were more easily killed than Japanese beetle grubs, a totally unexpected result (Table 3). For protection of lawns even the 2.2 and 4.5 kg/ha levels were considered sufficient for controlling at least 1 generation of both species. Dieldrin provided equally good control of both species. The 0.6 kg level gave satisfactory turf protection against both species for 1 generation. The Asiatic garden beetle averaging 10 and 28 grubs/0.1 m^2 was highly susceptible to both insecticides. All the grubs were killed in all except the 1.1-kg chlordane treatment, which provided 94% control.

DISCUSSION.—In laboratory tests both 1st- and 3rd-instar grubs of the European chafer were more difficult to kill than Japanese beetle grubs in the same stage. It required roughly twice as much of each

Table 1.—Relative susceptibility of 1st-instar grubs of the European chafer and the Japanese beetle to chlordane and dieldrin incorporated into mineral soil.

Insecticide and formulation	Ppm AI in soil[a]	Avg % mortality of 1st-instar grubs[b, c]			
		Indoor, constant 21°C		Outdoor fluctuating 14.5–25.5° C	
		EC	JB	EC	JB
Chlordane 10G	0.21	0	56	16	67
	.42	54	92	71	97
	.63	68	96	97	100
	.84	77	100	99	100
	1.04	88	99	94	100
	1.25	93	100	100	100
	1.46	96	100	100	100
Dieldrin 10G	.21	80	100	100	100
	.42	97	100	100	100
	.63	98	100	100	100
	.84	100	100	100	100
	1.04	100	100	100	100
No. alive in untreated controls					
Chlordane tests:		81	75	69	75
Dieldrin tests:		145	98	119	91

[a] 0.21–1.46 ppm equivalent to 0.2–1.4 lb/3-inch acre.
[b] E C = European chafer; J B = Japanese beetle.
[c] % mortality based on Abbott's formula.

Table 2.—Relative susceptibility of 3rd-instar grubs of the European chafter and the Japanese beetle to chlordane and dieldrin incorporated into mineral soil.

Insecticide and formulation	Ppm AI in soil[a]	Grub[b]	Avg % mortality in weeks exposure[c, d]				
			1	2	3	4	5
Chlordane 10G	1.04	EC	1	4	5	10	16
		JB	1	5	25	37	56
	2.09	EC	1	15	28	61	76
		JB	0	13	54	84	94
	4.18	EC	2	16	43	78	91
		JB	7	31	77	90	95
	8.36	EC	8	38	65	94	96
		JB	15	53	89	95	98
Dieldrin 10G	0.52	EC	3	0	1	8	9
		JB	0	8	11	23	56
	1.04	EC	3	2	4	10	26
		JB	18	25	56	82	92
	2.09	EC	1	0	9	26	50
		JB	12	21	46	69	83
	4.18	EC	4	24	51	80	95
		JB	0	21	66	92	100
			No. alive untreated controls				
		EC	159	152	151	149	144
		JB	137	130	126	121	114

[a] 0.52–8.36 ppm equivalent to 1–8 lb/3-in. acre.
[b] E C = European chafer; J B = Japanese beetle.
[c] Incubation at constant soil temperature of 21.5°C.
[d] % mortality based on Abbott's formula.

insecticide to produce comparable results in the chafer as in the Japanese beetle grubs.

A dissimilar situation occurred in the field plots. Chlordane was more effective against the chafer than the Japanese beetle, and dieldrin was equally effective against both species. These differing results between laboratory and field tests may reflect the influence of larval habits. In laboratory tests, the entire soil mass inhabited by the grubs had an equal concentration of the insecticides. In the field tests, insecticides were applied to the surface and were consequently concentrated near the surface. Therefore, the apparently greater susceptibility of European chafer grubs to chlordane in the field may indicate a difference in larval habits. If 1st-instar European chafers are more active than 1st-instar Japanese beetles and feed more often very near the soil surface, these seemingly contradictory results become more understandable.

When we consider that surface applications of chlordane at 10 lb/acre has given 92+% control and dieldrin at 3 lb/acre has given 100% control of European chafer grubs for 8 successive seasons with no indication of diminishing activity (Gambrell et al. 1968), it appears that the presently recommended rates of application are considerably in excess of requirements necessary to give 4–5 years' protection to turf. Surface applications of 2.2 kg chlordane and 0.6 kg dieldrin/ha gave adequate protection of turf against both species for a single season. It is reasonable to expect that 1.5 kg of the former and 1.1 kg of the latter (equivalent to 4 and 1 lb, respectively), will give sufficient grub control for turf protection for 3–4 seasons. Follow-up surveys in these plots for the next 2–3 years should give us a definite basis for the possibility of lowering the dosage schedule and thus partially resolve the current objections against the use of cyclodiene insecticides.

Tables 3.—Relative susceptibility of European chafer and Japanese beetle grubs to chlordane and dieldrin in field turf plots. Applications made June 17, surveyed October 1, ± 1 day. Webster Park, Webster, N. Y. 1968.

Insecticide and formulation	Kg AI /ha[a]	Av. no. grubs/ten 0.1 m² samples/plot[b, c]		% control[d]	
		E C	J B	E C	J B
Chlordane 10G	0.0	46.8 a	78.6 a		
	1.1	5.4 b	35.8 b	88	53
	2.2	5.0 b	13.8 c	88	82
	4.5	3.0 b	11.4 c	93	85
	9.0	0.0 b	0.6 c	100	99
Dieldrin 10G	.0	17.0 a	81.2 a		
	.6	.6 b	2.4 b	97	97
	1.1	.2 b	1.8 b	99	98
	2.2	.2 b	3.8 b	99	95
	3.4	.0 b	.0 b	100	100

[a] 0.6–9.0 kg/ha equivalent to 0.5–8.0 lb/acre.
[b] Means in each column for each insecticide not followed by same letter are significantly different at 1% level according to Duncan's multiple range test.
[c] E C = European chafer; J B = Japanese beetle.
[d] Based on Abbott's formula.

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